Subterranean Biology 55: 27–42 (2026)
doi: 10.3897/subtbiol.55.175751
https://subtbiol.pensoft.net

RESEARCH ARTICLE

A peer-reviewed open-access journal
Subterranean Biology
Published by The International Society for Subterranean Biology

The influence of subterranean habitats in the sensorial and brain morphology of hypogean and epigean Copionodontinae catfish (Siluriformes, Trichomycteridae)

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Academic editor: Fabio Stoch | Received 22 October 2025 | Accepted 23 January 2026 | Published 10 February 2026

https://zoobank.org/4C37C505-323C-41D4-A73F-0971C7789ED2

Citation: Rodovanski T, Bichuette ME, Cetra M, Mattox GMT (2026) The influence of subterranean habitats in the sensorial and brain morphology of hypogean and epigean Copionodontinae catfish (Siluriformes, Trichomycteridae). Subterranean Biology 55: 27–42. https://doi.org/10.3897/subtbiol.55.175751

Abstract

Brazilian cavefishes are among the richest troglobitic ichthyofauna worldwide including 13 genera. Most cavefish in the Neotropical region belong to Siluriformes. Trichomycteridae are highly diverse siluriforms, and the subfamily Copionodontinae are notably primitive in body shape, showing unique specializations to cave life. We studied the sensorial morphology referring to perception of the environment and processing of this information in four species *Copionodon pecten*, *C. lianae*, *Glaphyropoma spinosum*, and *G. rodriguesi*, of which *G. spinosum* is the only hypogean species, involved in perception of presenting some classical troglomorphisms such as reduction of eyes and body pigmentation, in addition to brain morphometry. Previous studies have demonstrated that troglobitic copionodontines are highly agonistic; therefore we measured the barbels, eyes and counted pores of the lateral canal system on the head. A PERMANOVA was performed to test for statistically significant differences in these morphological traits among the species groups. These similarities were significant among both genera and epigean and hypogean populations in terms of external morphometric/meristic and morphometry of the brain. *G. spinosum* has a longer telencephalon than the

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other studied species, a region related to olfaction, spatial memory and species-specific behavior such as social interactions and reproductive behavior, a region typically wider in the studied hypogean fishes. We detected troglomorphisms (character states related to isolation in subterranean habitats) in the laterosensory cephalic system of the whole group, such as the reduction of anterior canal segments in relation to other trichomycterids, indicating that all four species have exaptations to inhabit hypogean habitats. Although we could not confirm the existence of morphological differences between the two genera, the hypogean and epigean populations of these endemic Chapada Diamantina cave catfishes may exhibit morphological similarities.

Keywords
Brazil, cave fish, Chapada Diamantina, Neotropical fishes, sensory biology, troglomorphisms

Introduction

Brazilian cavefishes comprise one of the richest subterranean ichthyofauna on the planet, currently including 13 genera, seven families, and three orders with troglobitic (cave-restricted) species (Bichuette and Gallão 2021; Trajano 2021). This diversity is composed by many highly endemic monotypic genera, with morphological and ecological adaptations to the subterranean environment (Trajano et al. 2010; Bichuette and Gallão 2021). The majority of troglobitic fish belong to two different orders: Cypriniformes (carps, barbs and allies), a group of freshwater fishes that occur in all continents except South America and Antarctica, and Siluriformes (catfishes and armored catfishes), a group occurring worldwide including the Neotropical region (Bichuette and Gallão 2021). Brazilian cavefish are mostly Siluriformes, and Trichomycteridae are particularly abundant in caves. This family is a highly diversified neotropical group with a wide distribution, from Costa Rica to Patagonia in Argentina, found under different environmental conditions and occupying diverse habitats (de Pinna 1992). Trichomycteridae is currently classified into nine subfamilies: Copionodontinae, Glanapteryginae, Microcambevinae, Sarcoglanidinae, Stegophilinae, Trichogeninae, Trichomycterinae, Tridentinae, and Vandelliinae (Nelson 2016; Fricke et al. 2025).

The subfamily Copionodontinae occupies a basal position in the family, characterized by a plesiomorphic body shape, and represents a monophyletic group with some unequivocal synapomorphies such as the presence of exclusively spatulate dentition, absent in all other representatives of Trichomycteridae (de Pinna 1992; Bichuette et al. 2008). It is composed of two genera with seven species: *Copionodon* de Pinna, 1992 (*C. elysium* de Pinna, Burguer & Zanata, 2018, *C. exotatos* de Pinna, Abrahão, Reis & Zanata, 2018, *C. lianae* Campanario & de Pinna, 2000, *C. orthiocarinatus* de Pinna, 1992, and *C. pecten* de Pinna, 1992) and *Glaphyropoma* de Pinna, 1992 (*G. rodriguesi* de Pinna, 1992 and *G. spinosum* Bichuette, de Pinna & Trajano, 2008). The group was formally defined more than 30 years ago with the description of its first three species, definition of its genera and of Copionodontinae itself (de Pinna 1992). All remaining species were described after 2000 (Campanario and Pinna 2000; Bichuette et al. 2008; de Pinna et al. 2018a, 2018b).

Copionodontinae is endemic to the Chapada Diamantina karst area, Bahia, northeastern Brazil. They are found in the Rio Paraguaçu basin, inhabiting the upper reaches of fast-flowing streams running on rocky bottoms, which are interrupted leaving isolated pools in the dry season. The water in the upper reaches of Chapada Diamantina is rather cold and generally black; however, copionodontines are also found in a few clear water streams. Due to their agonistic behavior, they usually share the environment with few or no other species (Bichuette et al. 2008; Rantin and Bichuette 2019). Among Copionodontinae, *Glaphyropoma spinosum* is a subterranean species with some classical troglomorphisms, such as eye and pigmentation reduction (Bichuette et al. 2008). Its sister-species, *G. rodriguesi* is considered an epigean species occasionally found in caves (ME Bichuette, pers. obs.). Populations of *Copionodon pecten* and *C. orthiocarinatus* occur near the entrance zone and in epigean streams close to caves. In addition, there is a record of two copionodontine troglobitic species coexisting in caves: *G. spinosum* and one undescribed species of *Copionodon* (Rantin and Bichuette 2013, 2015, 2019; ME Bichuette, pers. obs.). In behavioral studies of these copionodontines associated with hypogean environments and surroundings, Rantin and Bichuette (2013, 2015, 2019) concluded that *G. spinosum* is more photophobic and cryptobiotic among three analyzed species (*G. spinosum*, *Copionodon pecten* and *C. orthiocarinatus*). In addition, cave copionodontines exhibit highly aggressive behavior (Rantin and Bichuette 2019).

Poulson (1963) observed that some morphological features of the brain and sensory system were related to troglomorphisms and specific behaviors of North American cave amblyopsids. The behavioral differences between the hypogean and epigean populations of Copionodontinae are significant and reflect distinct survival strategies (Rantin and Bichuette 2013, 2015). It remains to be tested if these differences can be related to some morphological aspect of these fishes, especially regarding its environmental perception and information processing.

We analyzed the brain morphology and sensory structures (barbels length, eye size and characteristics of the cephalic laterosensory system) in four Copionodontinae species: *Glaphyropoma spinosum* (troglobitic), *G. rodriguesi* (epigean), and the epigean *Copionodon pecten* and *C. lianae*, representatives of the sister lineage of *Glaphyropoma* (de Pinna 1992). These hypogean fishes show notable taxonomic and function diversity related to habitat. We also compared this information with the behavior specializations observed in *G. spinosum* and *C. pecten* (Rantin and Bichuette 2013).

Material and methods

We selected four species of Copionodontinae (Siluriformes, Trichomycteridae) (Fig. 1): one troglobitic and troglomorphic (*Glaphyropoma spinosum*), its sister-species and only congener as an epigean species (*G. rodriguesi*), and two species belonging to the outgroups of *Glaphyropoma* (*Copionodon pecten* and *C. lianae*). The studied specimens (Table 1) were preserved in 70% alcohol and deposited in the ichthyological collection of the Laboratório de Estudos Subterrâneos, Universidade Federal de São Carlos

(**LES**, **UFSCar**) and the Museu de Zoologia da Universidade de São Paulo (**MZUSP**). The catfishes were sampled in streams belonging to the Upper Rio Paraguaçu basin, Chapada Diamantina region, central Bahia state, Brazil. We examined 14 specimens of *Glaphyropoma spinosum*, three specimens of *G. rodriguesi*, 11 specimens of

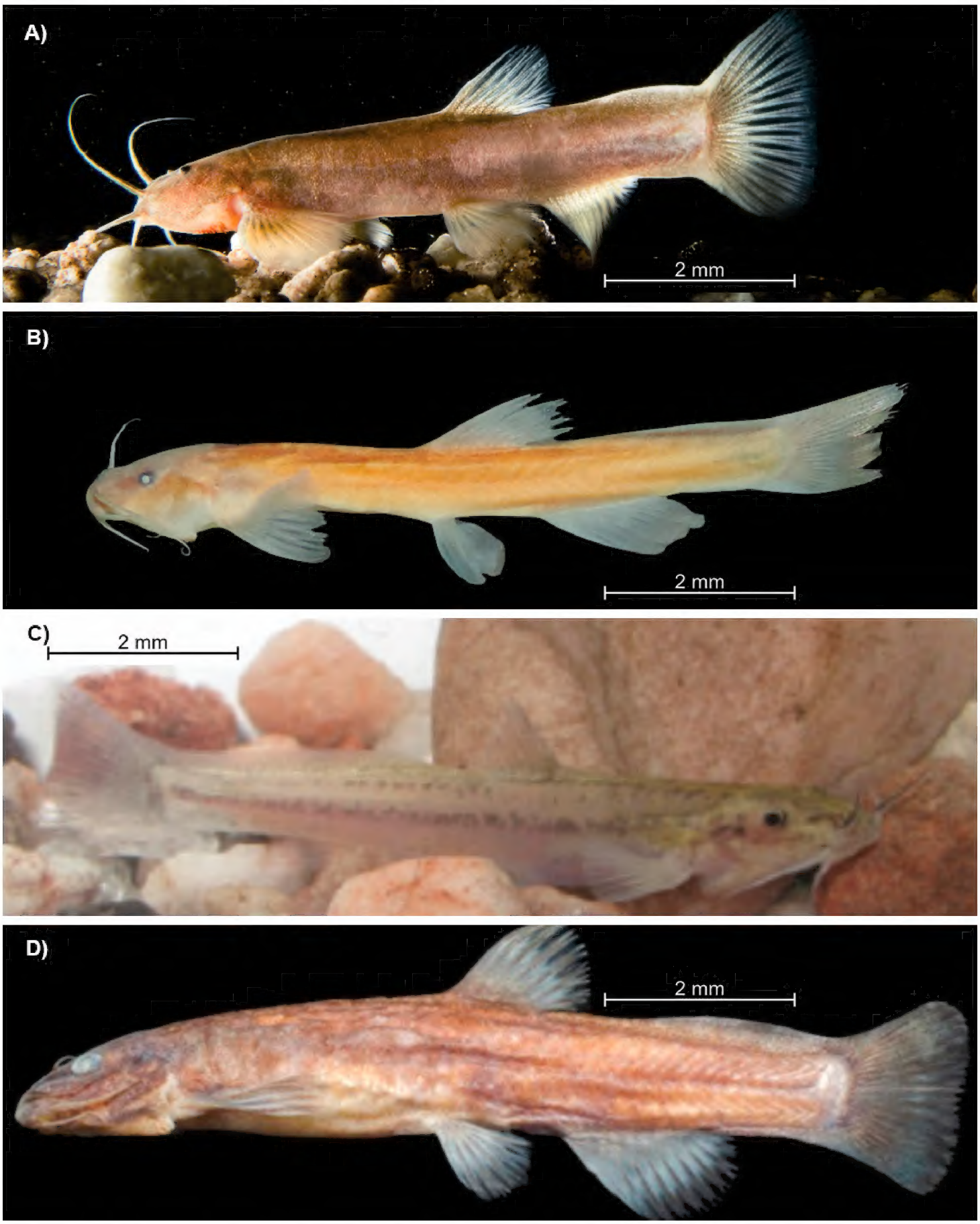


Figure 1. Live specimens of *Glaphyropoma spinosum* (**A**) and *Copionodon pecten* (**C**). Preserved specimens of *Glaphyropoma rodriguesi* (**B**) and *Copionodon lianae* (**D**). Authors from top to bottom: A. Gambarini; L. Ferreira; M. E. Bichuette; P. P Rizzato, respectively.

Table 1. List of examined material. All specimens were collected in Chapada Diamantina, Upper Rio Paraguaçu basin, central Bahia State, Brazil. N = 39. One asterisk denotes holotype and two asterisks indicate paratype.

Species	*N*	Locality	Habitat	Municipality
Glaphyropoma spinosum	8	Gruna dos Torras I (cave)	hypogean	Andaraí
G. spinosum	5	Gruna da Parede Vermelha (cave)	hypogean	Andaraí
G. spinosum	1	Gruna Ressurgência do Morro de Alvo (cave)	hypogean	Andaraí
G. rodriguesi	2	Gruna da Parede Vermelha (cave)	hypogean	Andaraí
G. rodriguesi *	1	Rio Mucugê	epigean	Mucugê
Copionodon pecten	11	Rio Coisa Boa	epigean	Andaraí
C. pecten **	1	Rio Mucugê	epigean	Mucugê
C. lianae	10	Rio Coisa Boa	epigean	Andaraí

Copionodon pecten, and 10 specimens of *C. lianae*. An additional specimen of *C. pecten* (a paratype) was examined exclusively for laterosensory canals. The limited number of specimens available for *G. rodriguesi* is due to the rarity of this species.

Collection permits were issued by Instituto Chico Mendes de Conservação da Biodiversidade (ICMBio 20165) and Commission of Animal Ethics of UFSCar (CEUA).

Measures were taken point to point with a digital caliper with the precision of one decimal case. These measures were based on Poulson (1963) and Trajano (1994) and included standard length (**SL**), head length (**HL**), eye diameter of both sides (**ROD** and **LOD**), interorbital width (**IW**), total area of nasal rosette (**ANR**), right nasal (**RNB**) and left nasal barbels (**LNB**), right maxillary 1 (**RMB1**) and left maxillary 1 barbels (**LMB1**), right maxillary 2 (**RMB2**), and left maxillary 2 (**LMB2**) barbels. All measures were calculated in relation to HL.

We then examined the cephalic laterosensory pores and canals following Rizzato and Bichuette (2017). In order to detail the laterosensory canals, we cleared and stained (c&s) specimens according to Taylor and van Dyke (1985). Nomenclature of the canals and pores follow Rizzato and Bichuette (2017). Before dissecting to examine the brain morphology, all specimens were photographed under a Zeiss Discovery V20 Stereoscope with Z-stacking autofocus attached to an Axiocam digital camera.

The dissection protocol followed the studies of Abrahão and Pupo (2014), further developed by subsequent studies (Abrahão and Shibatta 2015; Abrahão et al. 2018; Pupo and Britto 2018). We did this part of the study only with three of the four species included, with five specimens examined for each species (i.e., *Copionodon lianae*, *C. pecten* and *Glaphyropoma spinosum*). Due to the rarity of *G. rodriguesi* (n = 3, with one of them being the holotype), this species was not accessed in the brain morphometry part. We measured 14 brain features, the neuroanatomy nomenclature and the ten measurements were based on Trajano (1994) (Fig. 3): brain length (**EL**), mesencephalon length (**MSRL**) and width (**MSW**), telencephalon length (**TRL**) and two widths (**TTATW** and **TTPTW**), metencephalon length (**MTL**) and two widths (**MTATW** and **MTPTW**), and midbrain width. In addition, we estimated the volumes of each part of the encephalon (i.e., telencephalon [TV], mesencephalon [MSV] and

metencephalon [MTV]) by multiplying the height, length and width of each region. The two widths measured in the telencephalon and metencephalon correspond to the two transversals through the anterior third and base of the structure (i.e., the wider width of the region). Measures of the brain were taken with digital photographs using the above equipment with the aid of Zen Software. Most point to point measures (except heights) were calculated in relation to brain length. Heights were measured in the largest height possible of each region in order to calculate the volume.

To evaluate whether eye size, barbel length, and brain features differ among species groups, a Permutational Multivariate Analysis of Variance (PERMANOVA) was conducted. The analysis used a dissimilarity matrix based on Euclidean distances, calculated from measurements previously standardized by head length or brain length for external morphology and brain regions, respectively. Significance was determined through 999 permutations. The models included genus, habitat, and their interaction as explanatory variables. Additionally, non-metric multidimensional scaling (NMDS) plots were used to visualize morphological relationships among species. All analyses were performed in R (R Core Team 2024). Finally, we acknowledge the limitations of our analyses due to the partial overlap between the factors tested (e.g., the absence of hypogean *Copionodon* species).

Results

Eyes and barbel size

The comparative analysis of the four species included in this study demonstrates that both *Glaphyropoma spinosum* and *G. rodriguesi* have eye reductions or even asymmetry in the presence of the eyes, with a few specimens lacking one eye, a condition expected in hypogean species such as *G. spinosum*. Thus, we hypothesize that the asymmetry in the presence of the eyes is a plesiomorphic character of the genus rather than of a feature associated with the habitat in which each species inhabits, and that eventually favoured the colonization of hypogean habitats by *G. spinosum*. In other words, *Glaphyropoma* would have the tendency to have reduced eyes and this favoured the colonization of hypogean habitats by *G. spinosum*. Moreover, both species of *Glaphyropoma* have longer barbels (Table 2) and larger heads when compared to *Copionodon pecten* and *C. lianae*, features associated with troglomorphic characters (Fig. 2).

The PERMANOVA model testing the interaction between genera (*Copionodon* vs. *Glaphyropoma*) and habitat (epigean vs. hypogean) revealed significant differences in eye and barbel size among species groups (Pseudo-$F_{2,35}$ = 20.89, $p < 0.0001$). When factors were analyzed independently, both genera and habitat were significantly associated with this morphological variation. The genus accounted for more than half of the total variation (Pseudo-$F_{1,36}$ = 42.24, $p < 0.001$, $R^2 = 0.54$), while habitat also exerted a significant effect (Pseudo-$F_{1,36}$ = 21.55, $p < 0.001$, $R^2 = 0.37$) (Fig. 4). These results suggest a partial overlap in the explained variance, likely reflecting the inherent correlation between taxonomy and habitat specialization in the studied groups.

Table 2. Range of barbel length in the four species examined. Characters 1–8 are absolute and given in mm; characters 9–15 are proportion in relation to head length.

		Glaphyropoma spinosum	*G. rodriguesi*	*Copionodon pecten*	*C. lianae*
1	**Standard length**	32.2–71.9	47.4–57.2	37.9–52.5	38.3–50.7
2	**Head length**	6.6–13.8	8.6–11.4	5.8–9.8	7.0–9.2
3	**Right nasal barbel length**	8.2–23.0	9.9–22.6	3.2–4.4	3.9–5.4
4	**Left nasal barbel length**	9.0–23.1	10.5–23.0	3.5–4.4	3.9–5.4
5	**Right maxillary 1 barbel length**	7.8–23.3	10.5–24.2	5.4–6.9	4.5–7.5
6	**Left maxillary 1 barbel length**	8.3–23.5	11.1–24.8	5.4–7.1	5.7–7.3
7	**Right maxillary 2 barbel length**	3.7–12.1	6.8–10.5	2.6–4.8	3.3–4.5
8	**Left maxillary 2 barbel length**	3.5–12.6	6.5–9.4	2.5–3.7	3.4–4.3
Proportions of Head length					
9	**Right nasal barbel length**	110.1–168.1	114.7–198.3	0.0–58.9	47.7–71.4
10	**Left nasal barbel length**	110.0–178.5	143.2–201.4	37.2–59.6	47.8–71.7
12	**Right maxillary 1 barbel length**	98.4–173.2	147.0–221.9	57.5–98.3	64.1–87.6
13	**Left maxillary 1 barbel length**	115.2–170.8	144.7–217.3	57.5–110.1	0.0–84.9
14	**Right maxillary 2 barbel length**	0.0–95.7	92.2–93.3	28.8–67.9	40.7–56.3
15	**Left maxillary 2 barbel length**	49.4–102.8	80.9–81.9	28.4–49.5	42.4–56.6

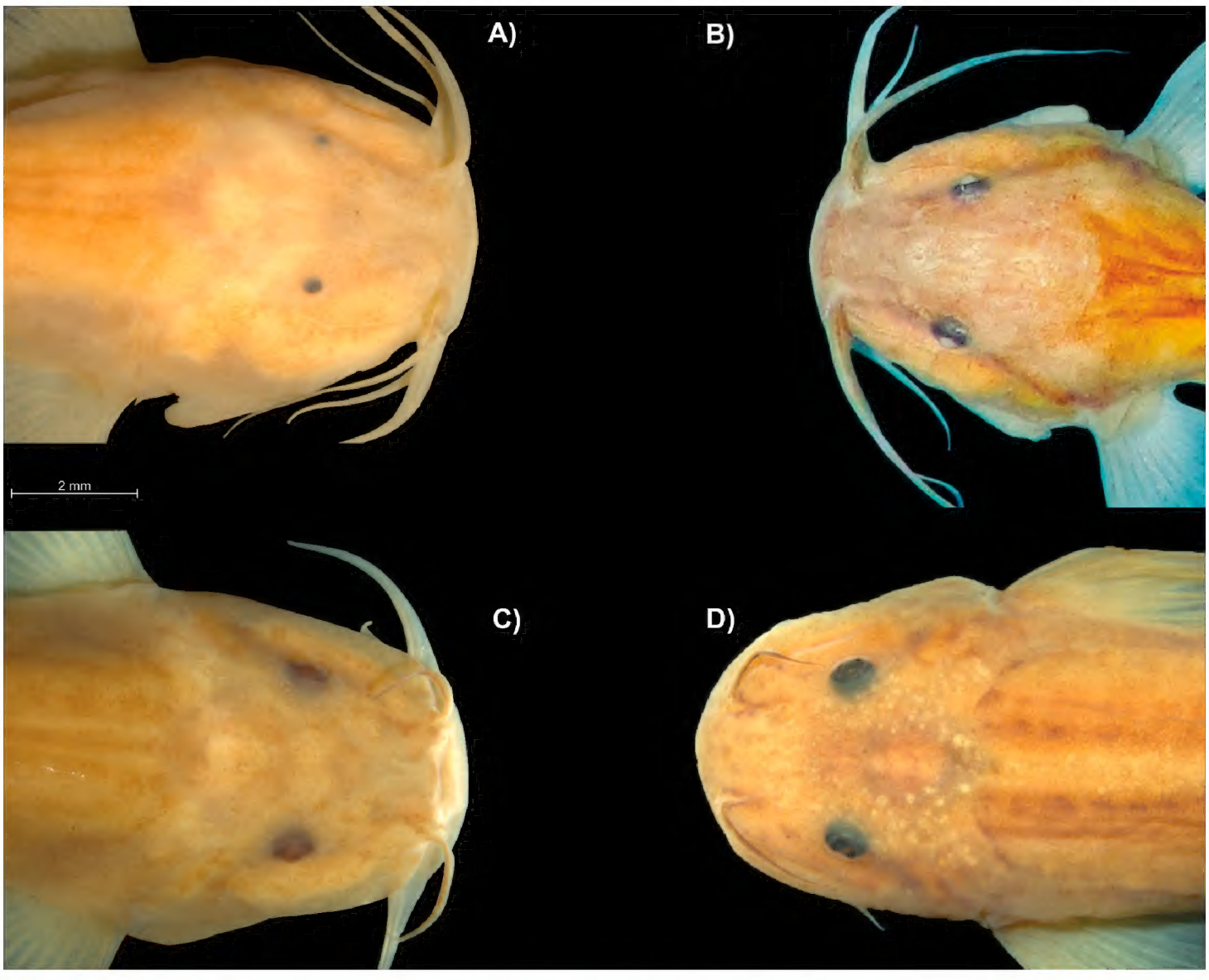


Figure 2. Dorsal views of the anterior portion of the body in the studied Copionodontinae species. **A** *Glaphyropoma spinosum* (52.9 mm SL) **B** *G. rodriguesi* (57.5 mm SL) **C** *Copionodon pecten* (54.4 mm SL) **D** *C. lianae* (44.9 mm SL).

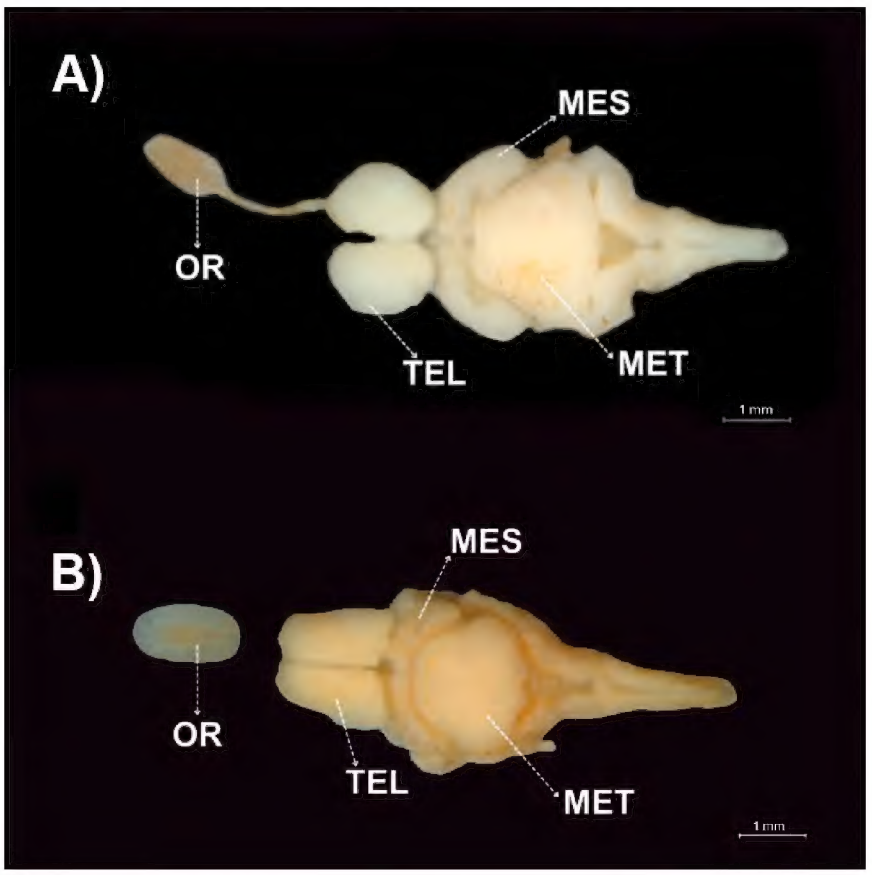


Figure 3. Dorsal view of the encephalon and olfactory rosette of **A** *Copionodon pecten*, 47.7 mm SL, and **B** *Glaphyropoma spinosum*, 55.2 mm SL. Abbreviations: MES, Mesencephalon; MET, Metencephalon, OR, Olfactory Rosette; TEL, Telencephalon. Olfactory nerve missing in (**B**).

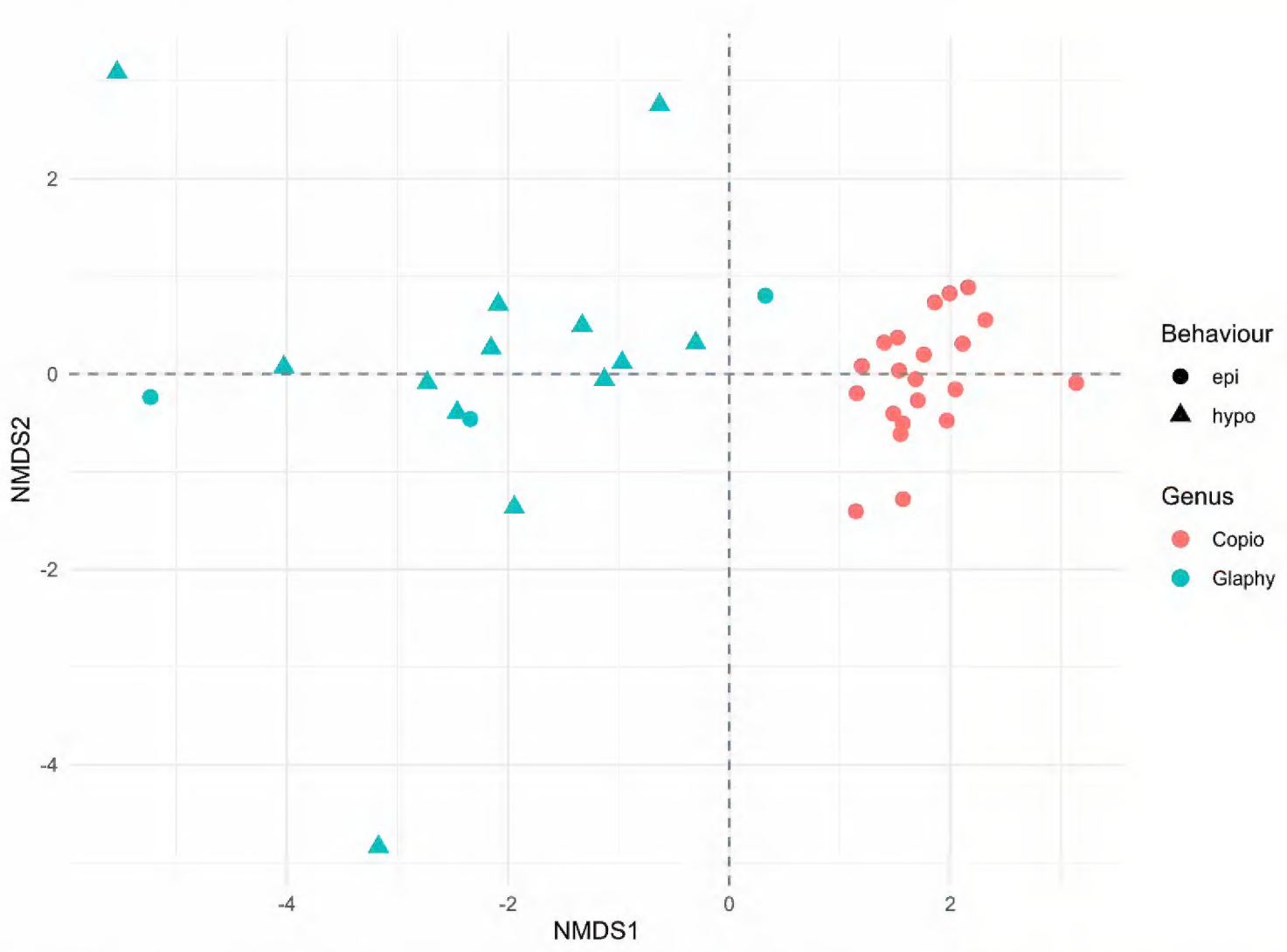


Figure 4. Non-metric multidimensional scaling (NMDS) ordination based on Euclidean distance of eye and barbel measurements. Colors represent the genera *Copionodon* (salmon) and *Glaphyropoma* (teal), while shapes indicate epigean (circle) and hypogean (triangles) habitats. Stress = 0.05.

Cephalic laterosensory system

The following canals were identified in the four clear and stained (c&s) specimens of the analyzed species: supraorbital (SO), infraorbital (IO), otic (OT), postotic (PO) and trunk (TR) (Figs 5, 6). The SO includes up to three canal segments (CS): anterior nasal, posterior nasal and frontal segments. The parietal branch was observed in a single c&s specimen of *Copionodon pecten* (n = 11) and in three specimens of *Glaphyropoma spinosum* (one of which c&s). The IO usually includes two canal segments: the antorbital CS and the infraorbital CS, although four segments were found in a single c&s specimen of *G. rodriguesi* and on the right side of a single c&s specimen of *C. pecten*. The OT, PO, and TP canals each included a single CS (Fig. 5).

The preopercular-mandibular canal was not accurately observed; thus, it was not included in the analyses.

Absence of canals is a tendency that has been described in subterranean fishes and is related to a troglomorphic condition (Rizzato and Bichuette 2017). This condition was observed in all four species. Furthermore, the asymmetry of the pores was observed in all specimens which was not expected for the epigean species.

Brain

Brain morphometry of *Glaphyropoma spinosum* differed from the two *Copionodon* species included in this study. In the former species, the relative telencephalon length (TRLr) is greater (Figs 3, 8).

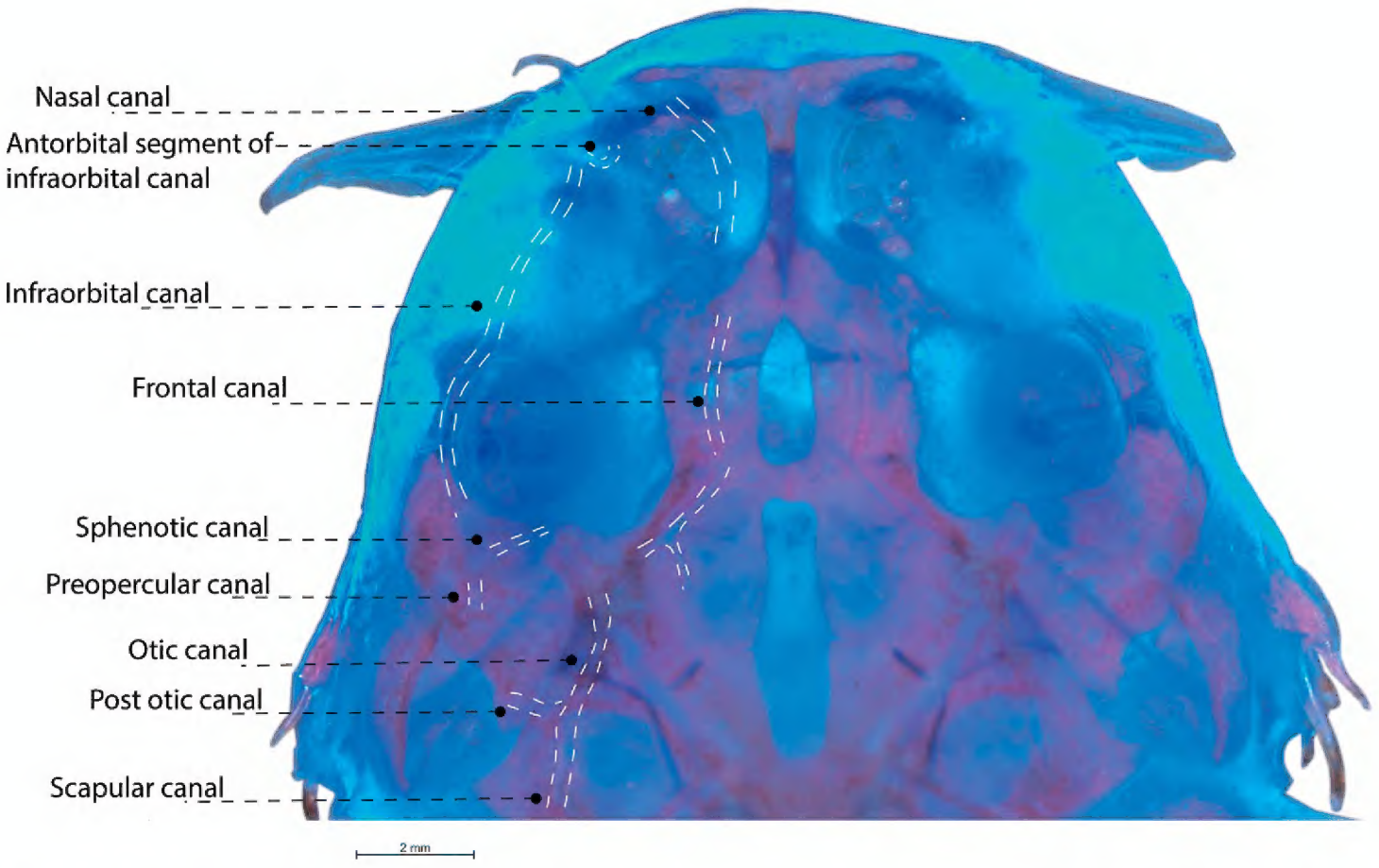
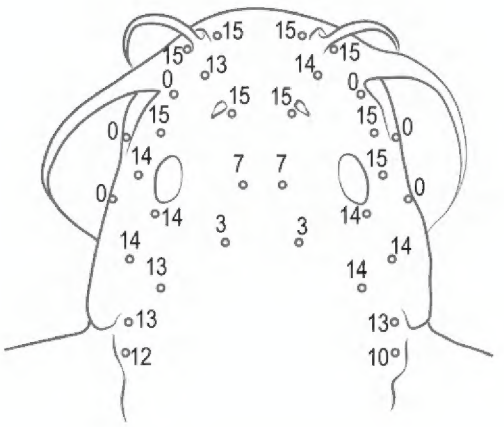
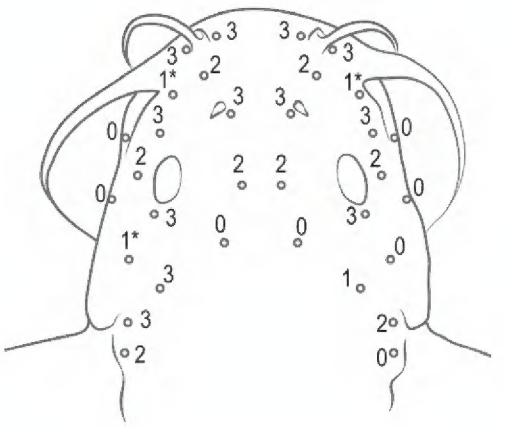


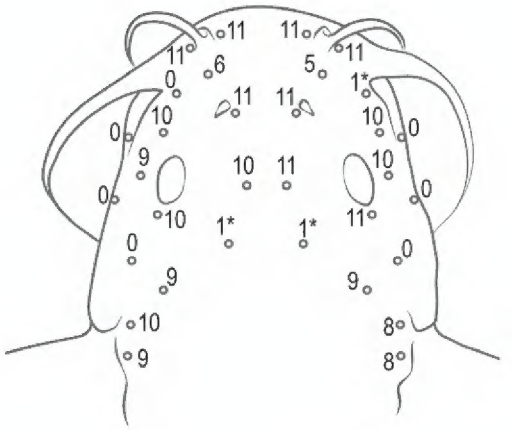
Figure 5. Dorsal view of the head of *Copionodon pecten* illustrating part of the cephalic laterosensory system (MZUSP 42462, 46.1 mm SL).



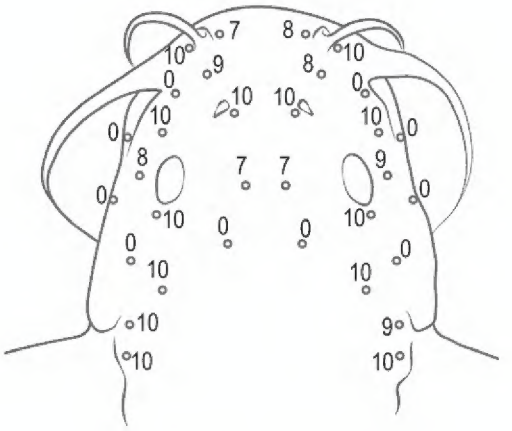
Glaphyropoma spinosum
N = 15



Glaphyropoma rodriguesi
N = 3



Copionodon pecten
N = 11



Copionodon lianae
N = 10

Figure 6. Variation in the number of pores of the cephalic laterosensory system in the analyzed species. The number beside each pore is the total number of specimens with that pore out of the total number of specimens examined for each species (N below each scheme). Asterisk beside number of count in *Copionodon pecten* represents single specimen examined with respective pore Author: P.P. Rizzato.

The PERMANOVA model indicated significant differences in brain volumes among the three species, explained approximately 32% of the total variance (Pseudo-$F_{2,12} = 2.80$, $p < 0.05$) (Fig. 9). The primary driver of this differentiation was telencephalon length (Fig. 8). However, it was not possible to isolate whether this variation stems from genus, habitat specialization, or an interaction of both. This limitation arises from the lack of hypogean *Copionodon* species in our dataset, resulting in confounded effects where variations attributed to both factors overlap.

Discussion

Results obtained through morphometric, meristic, and statistical analyses of the analyzed sensory structures in members of the catfish subfamily Copionodontinae revealed that the troglobitic species, when compared to epigean species, exhibit significant differences correlated with both factors studied (taxonomic and habitat). That is, most of the variation observed in *Glaphyropoma spinosum* (a troglobitic species) characters is also observed in *G. rodriguesi*, but also in *Copionodon pecten*, *C. lianae* (all three epigean species). As mentioned above, we were unable to differentiate between

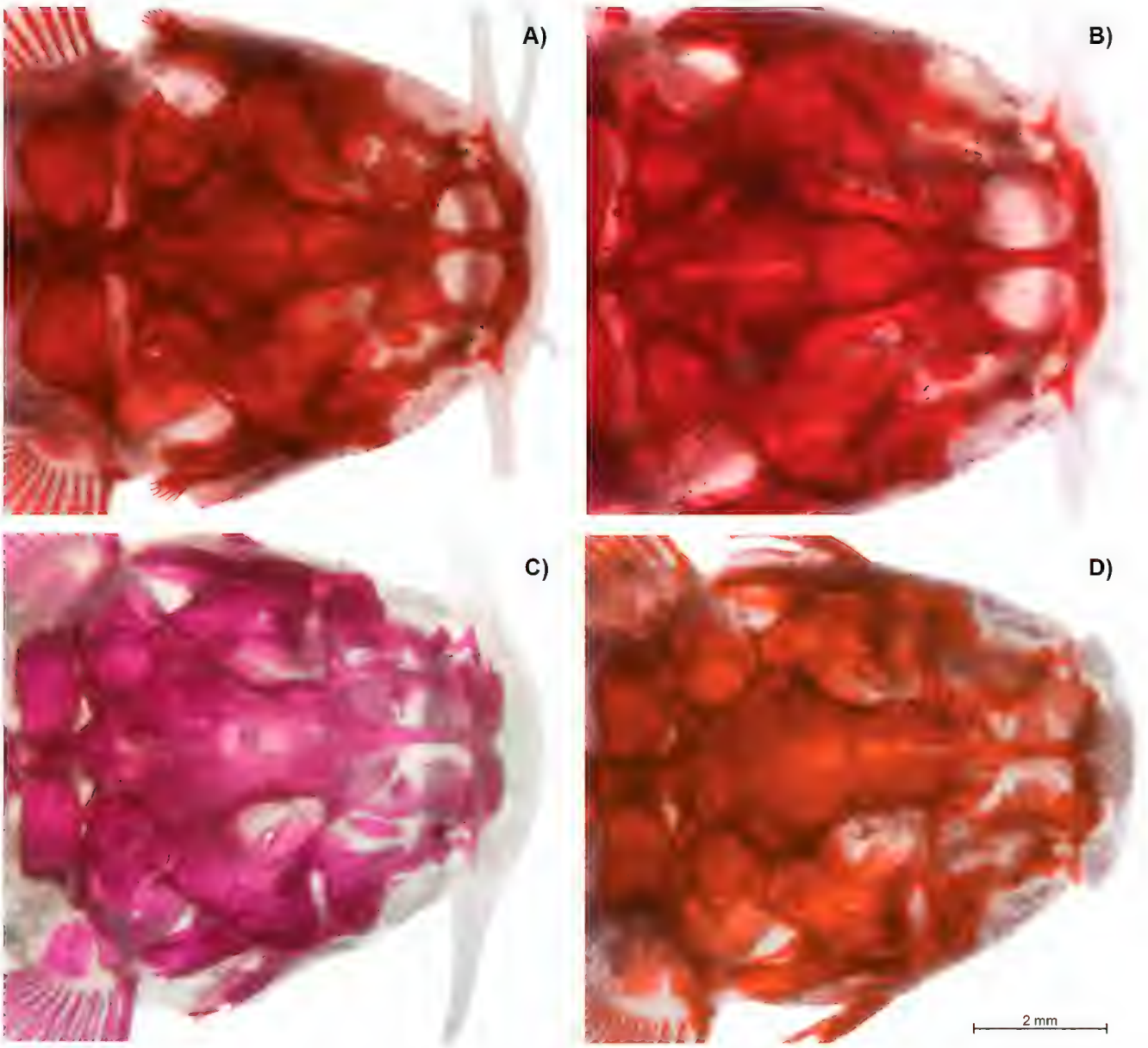


Figure 7. Details of the dorsal surface of the head of c&s specimens of **A** *Glaphyropoma spinosum*, **B** *G. rodriguesi*, **C** *Copionodon pecten* and **D** *C. lianae*, showing some of the lateral sensory canals examined herein.

the influence of one factor (taxonomic) and the other (habitat). This may be due to the fact that the analyzed *Copionodon* are epigean species, being nested in the taxonomy factor. In addition, the difference in brain morphometry could also be related to taxonomic factors, something that could not be tested in the present study because of the lack of brains of the epigean species of *Glaphyropoma*, *G. rodriguesi*.

There are many different sensorial structures on the anterior portion of the head that can vary among distinct fish groups. In Siluriformes, in addition to the eyes, there is chemoreception (i.e., barbels covered with taste buds used to feel the environment and savour food; Green and Zielinski 2013). Generally, there are three types of paired barbels in Trichomycteridae catfishes according to their position in the head: the nasal barbel near the nasal opening and two pairs of maxillary barbels associated with the gape of the mouth (Nelson et al. 2016). Such structures can vary depending on the taxonomic group [i.e., *Glaphyropoma* species have visually longer barbels (Table 2) than *Copionodon* species included herein, Fig. 2] and according to the habitat in which the animal lives and, in the case of cave catfishes, it is common to observe troglomorphisms (i.e., morphological characters commonly found in organisms of dark places such as caves). Morphological modifications generally interpreted as troglomorphisms

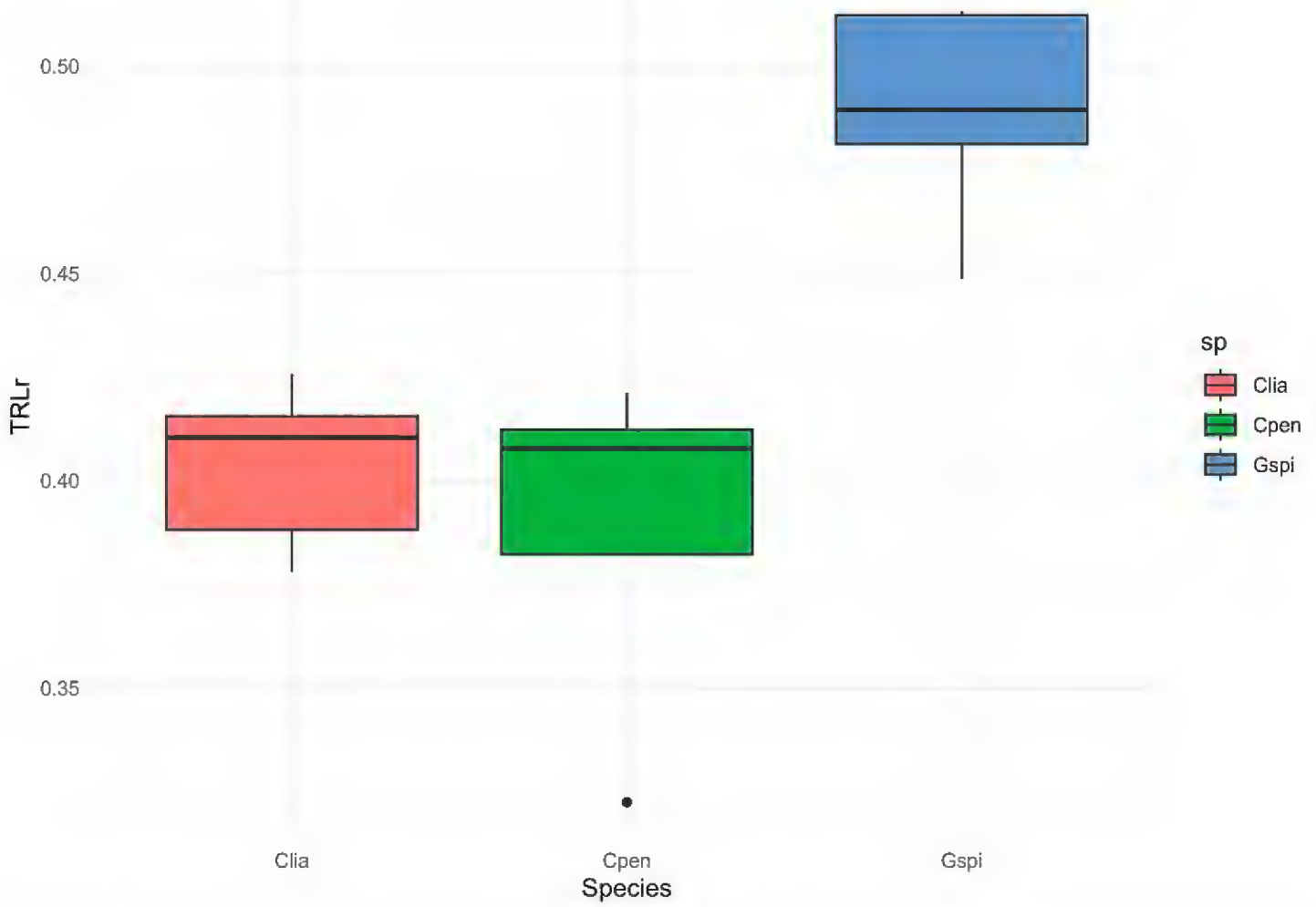


Figure 8. Boxplot of relative telencephalon length (TRLr) among *Copionodon lianae* (Clia, red), *C. pecten* (Cpen, green), and *Glaphyropoma spinosum* (Gspi, blue).

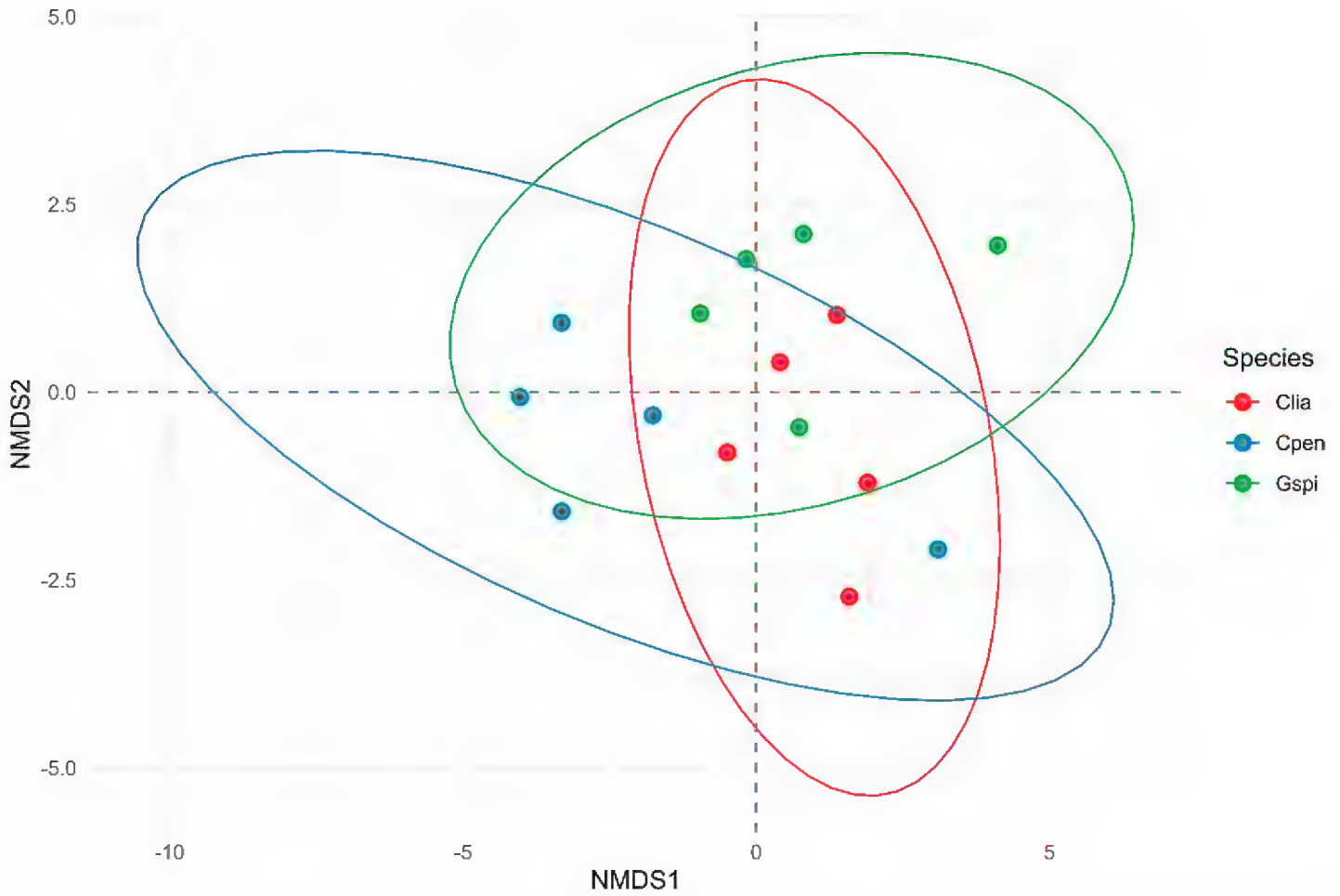


Figure 9. Non-metric multidimensional scaling (NMDS) ordination based on Euclidean distance of brain morphology data of *Copionodon lianae* (Clia, red), *C. pecten* (Cpen, blue), and *Glaphyropoma spinosum* (Gspi, green). Ellipses represent 95% confidence intervals for each species. Stress = 0.11.

are the reduction of the eye and body pigment, in some instances even completely lacking dark colour, as well as the increase of the surface with sensory units, like barbels (e.g., Trajano and Bichuette 2006; Bichuette et al. 2008; Trajano et al. 2010).

It was also noted that some traits in *Glaphyropoma spinosum* vary in their degree of troglomorphism, such as eye absence/asymmetry (e.g., loricariids – Secutti and Tajano 2009; heptapterids – Wilkens 2001) and the presence or absence of segments of canals in the lateral sensory system (e.g., trichomycterids – Rizzato and Bichuette 2017). This variability was also observed for behavior (Rantin and Bichuette 2013, 2015, 2019).

Troglobitic species may also exhibit a phenomenon called constructive traits, where certain structures are enhanced to compensate for the loss of a sense (e.g., olfaction vs. vision or telencephalon larger in hypogean species). For instance, the lateral line sensory system is often enhanced in fish (Poulson 1963; Trajano and Bichuette 2006). However, comparative data from the four studied species did not reveal any significant differences in the canal presence or pore count per canal. Significant brain differences are linked to the metrics of the telencephalon, a region associated with olfaction, spatial memory, and behavior (Trajano 1994; Liem et al. 2001). Previous studies have noted a trend toward larger telencephalons in troglobitic fish compared to epigean relatives (Trajano 1994). In terms of volume, this was also verified herein for *Glaphyropoma spinosum* (Fig. 9).

Morphometry of the brain of *Glaphyropoma spinosum* differed between the two species of *Copionodon* included in this study. Such features are expected in subterranean fishes because the former region is associated with olfactory perception and agonistic interactions. According to Trajano (1994), in hypogean fishes with reduced or absent eyes, there is a tradeoff between olfaction and vision in the dark environment of subterranean waters.

Therefore, even though it was not possible to identify significant differences in most portions of the brain, it is worth noting that there is a degree of troglomorphism in the brain of *Glaphyropoma spinosum* when compared to *Copionodon*. To be certain of this, the brains of *G. rodriguesi* specimens would be necessary. As for the mesencephalon, a region associated with vision (therefore expected to be small or less developed in species with small eyes), our results did not show a significant difference between the species and habitats.

Additionally, Rantin and Bichuette (2019) described agonistic behavioral patterns in two Copionodontinae species (epigean *Copionodon pecten* and hypogean *Glaphyropoma spinosum*) and concluded that both epigean and troglobitic species display this behavior. The authors showed that *G. spinosum* is highly aggressive, with a high frequency and complexity of aggressive components, including five new behavioral components for fishes in general. The determining factors for establishing dominance are body size, physical strength, and territoriality. Dominance is probably maintained by avoidance behavior by the subordinate and by continuous attacks by the dominant. They are highly intolerant to conspecifics, which may be a consequence of territoriality or feeding context. Two hypotheses were proposed by Rantin and Bichuette (2019) considering the agonistic behavior: (1) it is the maintenance of their ancestral behavior, plesiomorphic in relation to the remaining Trichomycteridae subfamilies, corroborated by phototactic and spontaneous behavior; (2) it is an autapomorphy for Copionodontinae, due to new evolutionary acquisitions. The epigean *C. pecten*, in contrast, is much

less aggressive, and its high tolerance to conspecifics and the absence of accentuated territoriality may be explained by food and/or space abundance in its natural habitats.

This suggests that, as hypothesis (1) proposed for agonistic behavior (as a plesiomorphic character state) indicates, the sensorial morphology follows a similar pattern. Furthermore, some traits are correlated to subterranean life, suggesting that some representatives of the Copionodontinae subfamily apparently show character-states to survive in a selective regime as the hypogean one.

Conclusion

In summary, this study aimed to verify whether there are morphological differences in the sensory structures of four species of the Copionodontinae, which correspond to the specialized behavior unique to the troglobitic species of the subfamily. It was possible to observe that, besides the statistically significant differences between both factors analyzed (genera and habitat), it is also possible to affirm that there are some troglomorphisms related to the brain and sensory canals, like elongated telencephalon in *Glaphyropoma spinosum* (the hypogean species) and absence/asymmetry of canals and pores in all species included herein. Due to the limited number of available specimens of *G. rodriguesi*, the only known epigean species of the genus, which occurs sporadically in caves, and because the single known specimen of the possible troglobitic species of *Copionodon* was not examined, it was not possible to determine whether the sensory troglomorphisms detected herein are related to taxonomic identity or habitat.

Author contributions

TR – data curation, formal analysis, investigation, writing original draft, writing review & editing. MEB – conceptualization, data curation, funding acquisition, resources, supervision, validation, writing original draft, writing review & editing. MC – formal analysis, writing original draft, writing review & editing. GMTM – conceptualization, data curation, funding acquisition, resources, supervision, validation, writing original draft, writing review & editing.

Acknowledgements

TR acknowledges PIBIC/CNPq (2023-2024) fellowship. GMTM and LISO were funded by FAPESP (Proc. 2017/01970-4). Fieldwork to collect the specimens was funded by FAPESP (Proc. 2008/05678-7) to MEB. The authors are grateful for discussions on the lateral line system with Pedro P. Rizzato (Departamento de Zoologia, Instituto de Biociências, Universidade de São Paulo) who also provided the drawings of Fig. 6. Eleonora Trajano and the editor provided helpful insights in an earlier version of the manuscript.

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